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QUASSIA EXTRACT AS A CONTACT INSECTICIDE

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INTRODUCTION

According to the literature, it appears that the extract from the Jamaica quassia wood (*Picrasma excelsa* Swz.) when properly extracted and applied is an efficient and satisfactory insecticide for the hop aphid (*Phorodon humuli* Schr.); but, owing to the fact that the active constituent of quassia wood is not toxic in the usual sense, authorities on insecticides are not yet agreed concerning the efficiency of quassia extract. Since this extract has never been used extensively upon other species of aphids, it is desirable to know whether or not it may be employed as a general insecticide for all aphids. Before being able to determine this point for aphids in general, it is first necessary to make a careful study of the economic methods of the extraction of quassia wood in order to determine what process assures the most thorough extraction of such constituents as are found by means of tests on aphids to be the toxic principles. It is further necessary to observe the physiological effects of this poisonous substance upon aphids. In this investigation, therefore, two chief objects have been kept in view: (1) To determine the efficiency of various extracts of quassia wood, and (2) to study the pharmacological effects of these extracts upon insects.

HISTORICAL REVIEW

I.—LITERATURE DEALING WITH QUASSIA AND QUASSIIN

According to the Ninth Decennial Revision of the United States Pharmacopœia, official quassia is derived from either *Picrasma excelsa* (Swz.) Planch. (family Simarubaceae), known commercially as Jamaica quassia, or from *Quassia amara* L. (family Simarubaceae), known commercially as Surinam quassia. According to the literature, there are a number of other plants which furnish wood with similar characteristics, whose active constituent is identical with, or similar to, quassiin, the bitter principle and main constituent of official quassia. These plants

are as follows: *Quassia simaruba*, *Quassia excelsa*, *Quassia polygama*, *Pitcarnia excelsa* s. *amara*, *Picraena excelsa*, *Picrasma quassioides*, *Picrasma eilantoides*, *Ailanthus excelsa*, *Simaruba amara*, *Simaruba cedron*, and *Simaruba versicolor*. There is hardly a question, however, that some of these are derived from the same source.¹

According to Flückiger and Hanbury (13),² quassia was introduced commercially into Europe about the middle of the eighteenth century. At first it was obtained entirely from *Quassia amara*, but later, owing to the great demand, it was obtained largely from *Picrasma excelsa*, the Jamaica quassia, a much larger tree than *Quassia amara*.

Dujardin and Égasse (6) state that *Simaruba amara* was introduced into France in 1713 as a remedy for dysentery. One of the earliest writers to mention quassia as an insecticide was Brande (3), who, in 1825, stated that it was an effectual stomach poison for flies when used in the form of an infusion sweetened with brown sugar.

As early as 1779 Paarmann (31) wrote a review on quassia and its uses. With respect to the anthelmintic properties of the bitter principle, he concurs in the opinion of others that its action on intestinal worms is due to a stimulation of the intestinal secretion which prevents their development rather than to any directly poisonous effect. He also performed some experiments on the extraction of quassin by various methods.

In 1794 Lindsay (21) published an account of *Quassia polygama* which he claimed had long been used in Jamaica as a useful medicine in "putrid fevers." All parts of the plant except the pulp of the fruit are bitter. At that time large quantities were being exported to England for use in the brewing of ale and porter.

In 1796 Trommsdorff (45) conducted a series of experiments from which he concluded that the best way to extract quassia is to soak the finely chipped wood for some time in cold water and then boil it three times, each time with 12 portions of fresh water. In 1811 Pfaff (34) compared the wood and bark of *Pitcarnia excelsa* s. *amara*, and concluded that the bark is much more bitter than the wood. He found that cold water extracts the bitter principle entirely and that if the

¹ Several of these names are synonyms; others were incorrectly given in the papers cited. *Picrasma excelsa* Planch. (1846) and *Picraena excelsa* Lindl. (1838) are names of the tree described originally as *Quassia excelsa* Swz., which is now referred to the genus *Aeschrion*, established by Vellozo at an earlier date (1827), and becomes *Aeschrion excelsa* (Swz.) Kuntze. *Quassia simaruba* L. f. is a synonym of *Simaruba amara* Aubl. *Quassia polygama* Lindsay is referred to *Aeschrion excelsa* (Swz.) Kuntze. "*Pitcarnia excelsa* s. *amara*," published by Pfaff, is an error in the paper cited. There is no genus *Pitcarnia*; *Pitcairnia*, for which it may have been intended, is the name of a bromeliaceous genus allied to the pineapple, having nothing in common with the family Simarubaceae, to which the quassia-yielding plants belong. The author may have intended the name for *Picraena excelsa*, and the supposed synonym *amara* for *Simaruba amara*. *Picrasma quassioides* Benn. is a valid name; *P. eilanthoides* (Bunge) Planch. (not *P. eilantoides* as given in the paper cited) is supposed to be a synonym of it. *Ailanthus excelsa* Roxb. is the valid name of an Asiatic tree delightfully aromatic and very different from our ill-smelling *Ailanthus glandulosa* Desf. *Simaruba cedron* is an erroneous name, intended for *Simaba cedron* Planch. *Simaruba versicolor* St. Hilaire is a valid name.—W. E. Safford, Economic Botanist, Bureau of Plant Industry.

² Reference is made by number to "Literature cited," p. 528-531.

material is "rubbed," the solvent action of the cold water is greater than that of either hot or boiling water, the high temperature in the case of the latter resulting in an oxidation and consequent insolubility of the bitter principle.

In 1822 Morin (25), working with the bark of *Quassia simaruba*, reported the presence of quassiin and a volatile oil having an odor of benzoin.

In 1829 Fechner (10) came to the conclusion that "rubbing" facilitated the extraction of quassiin from *Quassia excelsa* and that boiling is of no benefit. In 1835 Winckler (48) made an exhaustive study of *Quassia amara*, contributing much to the information already acquired concerning quassiin. He considered quassiin to be of a basic nature, but later investigators proved that it is not a true base. The experiments of Kellar (19) in the same year, while not conclusive, raised the question as to whether quassia contained an alkaloid. Two years later Wiggers (47) pointed out that quassiin was of a nonbasic character. He also outlined in general the properties of quassiin, including its solubility in water, which he found to be 0.45 part in 100. This seems to be much greater than the actual solubility, but his results may have been influenced by impurities, which he himself claimed will increase solubility. The substance obtained he called "quassit," the ending "it" being used to indicate its nonbasic character.

In 1858 Rochelder (38) recorded the fact that both *Simaruba amara* and *Simaruba cedron* contain a crystallizable bitter principle similar to quassiin. In 1868 Enders (9) asserted that quassia was used as a substitute for hops in brewing. In his study he found quassiin to be almost insoluble in water, readily soluble in alcohol and chloroform, and insoluble in ether. He concluded that the toxic principle was not a glucosid and found that it was precipitated by tannic acid.

In 1882 Christensen (4) obtained 12 gm. of what he considered pure quassiin from 18 kgm. of *Picraena excelsa*. This is equivalent to a solubility of 1 to 1,500. The solubility in warm water was found to be less than that in cold water.

In 1884 Oliveri and Denaro (26-29) undertook experiments to determine the molecular structure of the quassiin molecule. In 1889 Dymock and Warden (7) investigated *Picrasma quassioides*, a native of China and the subtropical Himalayas, and found the bark and wood to be as bitter as quassia. A crystallizable principle was isolated which resembled quassiin. An alcoholic extract gave positive reactions with alkaloidal reagents. No pharmacological effects were found. In 1890 Dymock, Warden, and Hooper (8) in their *Pharmacographia Indica* quote Stewart as stating that the wood of *Picrasma quassioides* was used in the Punjab to kill insects. In the same year Massute (23) published his researches on the chemical constituents of *Quassia amara* and

Picraena excelsa. He studied various methods of extracting quassia and "picrasmin," as he called the bitter constituent of *P. excelsa*. In 1891 Shimoyama and Hirano (40) described *Picrasma ailantoides* Planch. They found a crystalline principle in the bark which corresponded to quassia. Four years later Merck (24) separated a substance which he called "quassol" from impure quassia by solution in ether. Its melting point was found to be 149° to 150° C., while that of pure quassia is 210° to 211°. It is further distinguished from quassia by its tastelessness. In the same year Hooper (16) obtained from *Ailanthus excelsa* an extremely bitter substance resembling quassia. According to Dragen-dorff (5), *Simaruba excelsa*, a Brazilian species, is used as a remedy for intestinal worms and for skin parasites.

According to the preceding review the principal constituents of quassia are quassia, picrasmin, quassol, an alkaloid (?), a volatile oil (?), resin, mucilage, and pectin. The constitutions of the first three are not definitely known, and the actual presence of the volatile oil and alkaloid has not been definitely established.

2.—LITERATURE DEALING WITH QUASSIA EXTRACT AS AN INSECTICIDE

It is reported that quassia extract has been used as an insecticide in Europe for many years, but the earliest authentic record found by the writers occurs in 1885. On this date Ormerod (30) reports that the hop growers in England found quassia extract efficient upon the hop aphid. The proportion of ingredients used was generally 6 pounds of quassia chips and 3 pounds of soft soap to 100 gallons of water.

Alwood (1) prepared a decoction of quassia by using 1 pound of chips to 2 gallons of water. He says:

Applied pure, it killed the lice (hop aphid) effectually where they were reached, but it will not spread. Only those under drops were killed. Diluted once it was still quite effective, but could not be used with any thoroughness. I do not consider this a practical remedy when used alone.

Smith (41, 42, 43) prepared a strong decoction of quassia and applied it externally and internally to rose chafers (*Macrodactylus subspinosus* Fab.). He claims that it was ineffective, regardless of how it was applied.

Riley and Howard (36, 37), basing their deduction upon the results of Alwood's experiments (1), report that quassia extract when used alone is greatly inferior to well-prepared kerosene emulsion for hop aphids.

Koebele (20) sprayed hop aphids on prune trees in the States of Oregon and Washington with a solution prepared in the proportion of 6 pounds of quassia chips and 3 pounds of soap to 100 gallons of water. He says:

The numerous ants attending the Aphidids were not destroyed by this wash, and they carried off the lice not destroyed by the application the following day, leaving the immature lice dead upon the leaves. The action of the quassia is very slow and considerable time elapses before the lice are all destroyed.

Washburn (46) reports that a spray solution consisting of quassia extract and soap, barring the expense of the chips and the time consumed in the extraction, is recommended for the hop aphid in Oregon.

Gould (14) ascertained that a quassia extract solution not containing soap was inefficient for the San José scale on pear trees.

Fletcher (11, 12) reports that the decoction prepared in the proportion of 78 pounds of quassia chips and 7 pounds of whale-oil soap to 100 gallons of water is the standard remedy for the hop aphid in Ontario, and also that it has given most satisfactory results against other aphids with no injury to the foliage of the trees treated.

Howard (17), discussing the experiments of Celli and Casagrandi, who determined that the fumes from quassia wood kill aerial mosquitoes, summarizes the conclusions of the Italian authors as follows:

It is, however, to be noted that for these odors, fumes, or gases to exercise their culicidal action they must fill or saturate the whole ambient; otherwise they produce only apparent death, or at most only a culicifugal action, which sometimes in houses may be useful in protecting man from being bitten by mosquitoes.

Piper (35) reports that a decoction prepared in the proportion of 28 pounds of quassia chips and 7 pounds of whale-oil soap to 100 gallons of water is used almost exclusively for the hop aphid. He says:

It is quite as effective against other species of Aphid. The whale-oil soap without the quassia is of somewhat less efficiency.

Henderson (15) determined that a strong decoction of quassia without soap was ineffectual upon the apple aphid in Idaho, but when soap was added to it and the mixture was then diluted and applied warm the result was that nearly all of the aphids were killed.

Theobald (44), writing about the apple sucker (*Psylla mali* Schm.) in England, says:

The only preventive we find of any use is spraying with quassia and soft soap as soon as the buds commence to swell and the larvæ are seen to be coming from the eggs. We usually use 6 or 8 pounds of soft soap and 8 pounds of boiled quassia chips to the 100 gallons of soft water.

Boucart (2, p. 376), compiling formulas and results obtained therefrom by several authors, cites six insecticides having a quassia basis. These are variously concocted, some being decoctions and others infusions, but each one contains soap, and, furthermore, one contains alcohol, one petroleum emulsion, and one carbolic acid. The originator of the one containing petroleum emulsion recommends it for destroying various caterpillars infesting fruit trees. The authors of the formulas containing only quassia extract, soap, and water say that these insecticides kill *Cochylis ambiguella* Hübn. (cochylis of the vine), the hop aphid, wheat aphid, green aphids, woolly aphid, peach aphid, gooseberry aphid, *Phytoptus ribis* W., and *Phytocoris militaris* Westw. (orchid bug).

Parker (32), in the laboratory sprayed branches of prune trees bearing prune aphids (*Hyalopectus pruni* Fab.) with a solution made by extracting

5.33 ounces of quassia chips with 2 quarts of water; 92 per cent of these aphids died. Other branches of prune trees were sprayed with a solution prepared in the proportion of 7 pounds of quassia chips to 250 gallons of water; 96 per cent of these aphids were killed. Parker ascertained that variously concocted formulas consisting of quassia extract and soap solution are effective upon the hop aphid (*Phorodon humuli* Schr.) in the field. He believes that the solution kills only by coming in contact with the insects. The same author (33, p. 6), in the laboratory and in the field sprayed hop aphids and prune aphids with three different formulas in the proportion of ingredients as follows: (1) 0.4 gm. of quassiin to 2,000 c. c. of water with whale-oil soap; (2) 0.4 gm. of quassiin to 2,000 c. c. of water with soap bark; and (3) 0.4 gm. of nicotine sulphate to 2,000 c. c. of water with soap bark. He concluded that the formulas containing quassiin were almost as effective as that containing nicotine sulphate. He did not test the effects of the quassiin solution upon other insects, but believes that it will prove effective elsewhere if used in proportions corresponding to the amounts of nicotine sulphate that are known to be effective.

METHODS OF PREPARATION AND EFFECTIVENESS OF QUASSIA EXTRACTS¹

An investigation of the effectiveness of a substance as an insecticide may be divided, as a rule, into two distinct phases: (1) The preparation and preliminary testing in the laboratory of various extracts obtained from the substance; and (2) the testing under outside or field conditions of those solutions containing the extracts that have proved efficient in the laboratory.

I.—METHODS OF EXTRACTING QUASSIA CHIPS

The bitter principle, quassiin, which is considered to be the active constituent in quassia sprays, is claimed to be only slightly soluble in water. It is important, therefore, to know just what method of extraction is likely to insure the greatest quantity of this constituent. A review of the literature reveals a wide difference in the results obtained in extracting quassiin from the chips. The solubility of the quassiin, as found by various investigators, appears to vary greatly, and, owing to the confusion existing with respect to the practical methods of extracting this substance, it seems advisable to include in the present investigation some experiments to determine the value of the various methods.

(a) QUANTITIES OF EXTRACT REMOVED BY SUCCESSIVE EXTRACTION

On the market, quassia chips vary greatly in size. A bag of this material is likely to contain pieces varying in size from several inches in length to very small fragments and sawdust. Since even the average-sized chips are quite large, it was thought probable that they could be

¹ The word "extract" throughout this paper means the solid material removed by a solvent.

extracted a number of times, and an effective extract obtained in each case. Consequently 10 gm. of medium-sized chips were extracted for 2 hours with 500 c. c. of distilled water. This mixture was then filtered and the filtrate evaporated to determine the quantity of extract obtained. The same chips were extracted successively six times by using the same amount of fresh distilled water each time, and the quantity of extract was determined after each extraction. The experiment was then repeated, the time of extraction in each case being 24, instead of 2 hours. The results are presented in Table I. In this and the other tables the quantity of extract does not necessarily mean the quantity of quassiin.

TABLE I.—*Quantity and percentage of extract dissolved by successive extractions of 10 gm. of quassia chips with 500 c. c. of distilled water for 2 and 24 hours, respectively*

Extraction No.	Quantity of extract obtained.				Extraction No.	Quantity of extract obtained.			
	Chips extracted 2 hours.		Chips extracted 24 hours.			Chips extracted 2 hours.		Chips extracted 24 hours.	
	Gm.	Per cent.	Gm.	Per cent.		Gm.	Per cent.	Gm.	Per cent.
1.....	0.1035	1.03	0.1335	1.53	4.....	0.015	0.15	0.0757	0.76
2.....	.0255	.25	.0275	.27	5.....	.0075	.075	.0145	.14
3.....	.0215	.21	.0197	.20	6.....	.0005	.005

It will be noted that after five extractions of two hours each, practically nothing further is extracted. The total extract in the five portions of 500 c. c. each is 0.1730 gm. When the chips were extracted for 24 hours, the total extract in the five portions of 500 c. c. each was 0.2309 gm., or 0.0579 gm. more than in the first case. It is evident from this that a relatively long period of soaking is essential in order to get the maximum quantity of quassiin in solution. It was observed that the extracts in all cases were bitter and the residues from the final extractions, although much fainter in taste, were still distinctly bitter. Figure 1 shows graphically the relative quantity of extract obtained in each case.

(b) EFFECT OF BOILING THE CHIPS

To determine the effect of boiling on the quantity of extract which may be dissolved from the chips, 10 gm. were boiled under a reflux condenser in 500 c. c. of distilled water for various periods of time. After cooling and filtering, the filtrate was evaporated and the quantity of extract was determined. Table II gives the results.

TABLE II.—*Quantity and percentage of extract obtained by boiling 10 gm. of quassia chips in 500 c. c. of distilled water for various periods*

Length of time chips were soaked.		Extract obtained.		Length of time chips were soaked.		Extract obtained.	
Hours.		Gm.	Per cent.	Hours.		Gm.	Per cent.
½.....		0.1467	1.46	8.....		0.2765	2.76
1.....		.1872	1.87	16.....		.2747	2.74
2.....		.195	1.95	24.....		.3327	3.32
4.....		.2435	2.43				

It would appear that boiling for one-half hour extracts as much material from the chips as cold maceration for 24 hours. These boiled extracts are all more highly colored than the cold macerated extracts, and it is to be expected that substances other than those found in the macerated extracts are dissolved from the wood under the influence of the higher temperature. Figure 2 illustrates graphically the results given in Table II. It will be observed that comparatively little extract

is obtained after four hours' boiling; therefore it would hardly be economical to boil the chips longer than four or five hours.

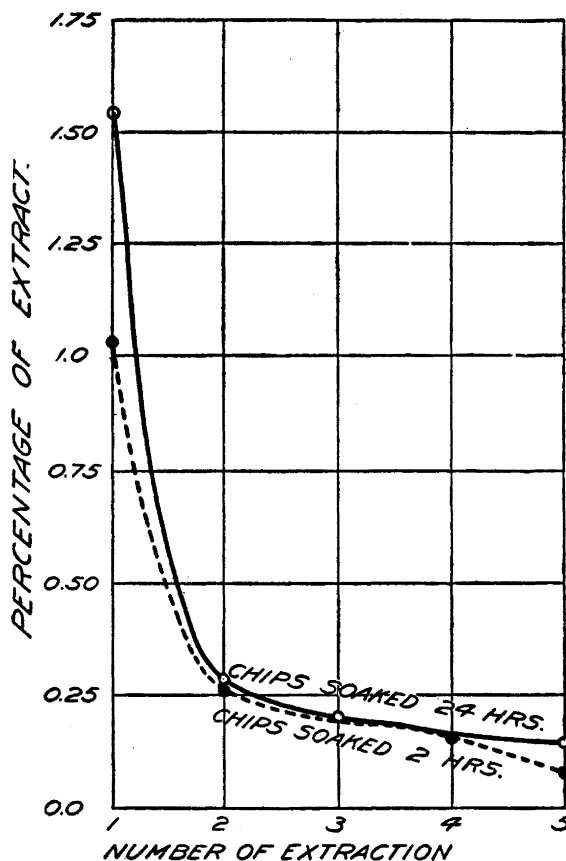


FIG. 1.—Graph showing the percentage of extract obtained by repeated soaking of 10 gm. of quassia chips with 500 c. c. of water for 2 and 24 hours, respectively.

pieces 6 to 8 cm. long and 1 to 2 cm. wide, to a No. 200 powder¹ In each case 10 gm. of the wood were macerated for 24 hours in 500 c. c. of distilled water. After filtering the mixture and adding enough water to make 500 c. c., the entire quantity was evaporated and the quantity of extract was determined. A second extraction was made in each case. Table III is a tabulation of the results obtained.

¹ A No. 200 powder is one that will pass through a sieve having 200 meshes to the inch.

(c) INFLUENCE OF SIZE OF CHIPS ON EXTRACTION

Whenever substances are extracted by maceration, the state of subdivision or comminution of the substance is of great importance. As a rule the finer the subdivision the more thorough is the action of the solvent. To determine whether the reduction of quassia wood to fine powder would materially increase the quantity of extract, a quantity of the wood was separated into a series of portions, varying in size from

TABLE III.—*Influence of size of chips on the quantity of extract obtained by macerating 10 gm. of quassia chips in 500 c.c. of distilled water for 24 hours*

Sample No.	Chips. Size of chips.	Quantity of extract obtained.				
		First extraction.		Second extraction.		Total.
		Gm.	P. ct.	Gm.	P. ct.	P. ct.
1	6 to 8 cm. long and 1 to 2 cm. wide. . .	0.0655	0.65	0.0260	0.26	0.910
2	4 to 5 cm. long and 1 to 1½ cm. wide. .	.0870	.87	.0280	.28	1.15
3	2 to 3 cm. long and ¾ to 1¼ cm. wide. .	.1257	1.26	.0295	.29	1.55
4	15 to 25 mm. long and 5 to 10 mm. wide. . .	.1147	1.15	.0217	.22	1.37
5	7 to 13 mm. long and 3 to 5 mm. wide. .	.1395	1.39	.0252	.25	1.64
6	No. 5 powder.1700	1.70	.0267	.26	1.96
7	No. 10 powder.1580	1.58	.0365	.36	1.94
8	No. 20 to 40 powder.1570	1.57	.0397	.40	1.97
9	No. 40 to 60 powder.1930	1.93	.0455	.45	2.38
10	No. 60 to 100 powder.2370	2.37	.0580	.58	2.95
11	No. 100 to 200 powder.2625	2.62	.0775	.77	3.39
12	No. 200 powder and finer.2930	2.83	.0910	.91	3.74

It will be noted that the quantity of extract dissolved by the water is proportional to the state of subdivision of the chips. This is especially true with regard to the first extraction. In figure 3, which shows graphically the relationship of the size of the chips to the quantity of extract obtained, it will be noted that relatively little extract is removed the second time. It is evident that a 24-hour maceration will extract from the chips such a percentage of the total water-soluble matter that a second extraction would be unprofitable. Reference to figure 1 will illustrate this further. It is noted that

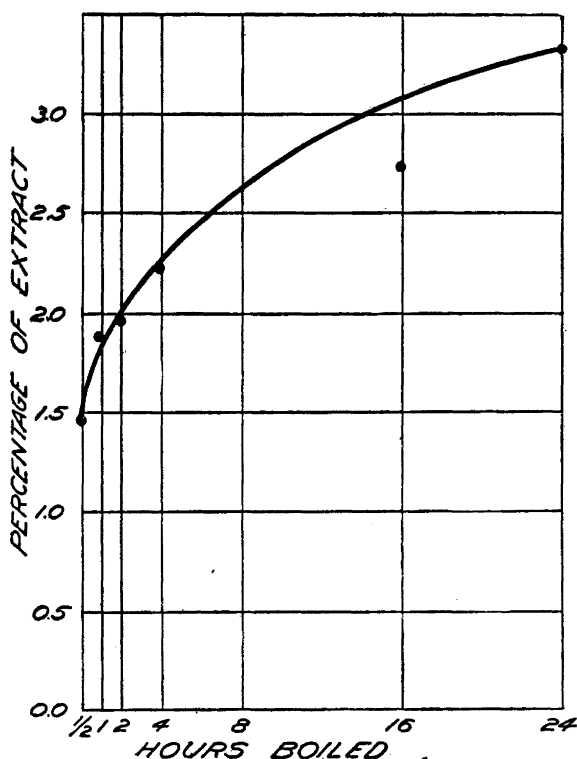


FIG. 2.—Graph showing the percentage of extract obtained by boiling 10 gm. of quassia chips with 500 c. c. of water for ½, 1, 2, 4, 8, 16, and 24 hours, respectively.

the second extraction removes only one-sixth as much material as the first.

Quassia wood is difficult material to get into a fine state of comminution. The coarser subdivision can be obtained by shredding and the next smaller division can be effected by means of burr mills. The fine powders, however, can only be secured by continued action of a pebble mill or a chaser mill. The expense of reducing the wood to fine powder would therefore be too great to make its use in that form economical. When the cost of grinding is taken into consideration, it seems that the most economical form in which to use quassia wood is that

corresponding to No. 5 to 7 in Table III. This material is about the size of coarse sawdust.

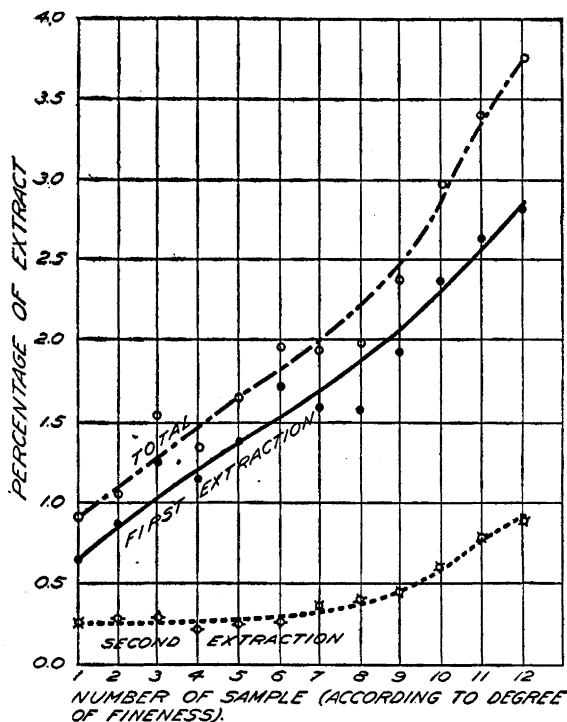


FIG. 3.—Graph showing the influence of the state of fineness of the quassia chips upon the percentage of extract obtained when extracting 10 gm. with 500 c.c. of water for 24 hours.

(d) INFLUENCE OF QUANTITY OF WATER USED

In making spray solutions from quassia chips it is a question whether it is more expedient to soak the necessary quantity of chips in the entire quantity of water or to soak them in a small quantity of water and later dilute the filtrate to the quantity of solution wanted. The latter method is, as a rule, the more convenient one in large operations, because the chips can be soaked in a suitable barrel and later the

dilution can be made in the spray tank. On the other hand, the first method, owing to the slight solubility of quassia in water, would appear to insure the more thorough extraction. This method, however, involves the use of a soaking tank large enough to hold the entire volume of spray solution. In the following experiment an attempt was made to determine the effect of the volume of water used on the total quantity of extract dissolved from the chips.

Four portions of 10 gm. each of medium-sized chips were macerated for 24 hours, in 250, 1,000, 2,000, and 3,000 c.c. of distilled water, respectively. After filtering, 200 c.c. of each filtrate were evaporated,

and the amount of extract was determined. The results are embodied in Table IV.

TABLE IV.—*Effect of the quantity of water used on the total quantity of extract obtained from 10 gm. of quassia chips macerated 24 hours*

Experiment No.	Quantity of water used as solvent.	Quantity of extract in 200 c. c. of filtrate.	Total quantity of extract in spray solution if sufficient water were added to make 3,000 c. c.
	C. c.	Gm.	Gm.
1.....	250	0.1054	0.1317
2.....	1,000	.0286	.1430
3.....	2,000	.0155	.1550
4.....	3,000	.0116	.1740

It may be well to explain the above table with a practical example. If it were desired to make a 3,000-c. c. spray solution with 10 gm. of quassia chips, either of the two following methods might be used: (1) The chips should be soaked for 24 hours in a small quantity of water (for example, 250 c. c.) , and after filtering the mixture sufficient water added to make 3,000 c. c.; or (2) they should be soaked in the full quantity of water (3,000 c. c.) for 24 hours and the mixture then filtered. From the table it will be seen that in experiment 1, in which 250 c. c. of water were used as a solvent, 200 c. c. of filtrate yielded 0.1054 gm. of extract, while in experiment 4, in which 3,000 c. c. of water were used as a solvent, 200 c. c. of filtrate yielded only 0.0116 gm. of extract. However, upon diluting No. 1 with sufficient water to make 3,000 c. c., the total extract in the spray solution was 0.1317 gm., while the total extract in the 3,000 c. c. No. 4 is 0.1740 gm. It is seen, therefore, that by using the entire quantity of water to extract the chips, 32.1 per cent more extract is obtained than by extracting in a small quantity of water and subsequently diluting. While, of course, it is to be expected that a considerable percentage of the water-soluble extract is not quassiin, it may safely be assumed that extracting with the total quantity of water assures a greater percentage of quassiin in the spray solution.

2.—EXPERIMENTAL TESTS FOR SELECTION OF EFFECTIVE FORMULAS

In the preceding pages the quantitative determinations show the following: (1) Chips soaked for 2 hours in water yield during the first extraction four times as much extract as during the second extraction; but chips soaked for 24 hours yield during the first extraction six times as much extract as during the second extraction. (2) Chips boiled longer than four hours yield but little more extract than those boiled for this period, and the quantity obtained is about one and one-half times that

obtained when chips are soaked for 24 hours. (3) The smaller the chips and the finer the powder, the greater is the quantity of extract removed. And (4) the larger the volume of water used as a solvent, the greater is the quantity of extract removed—that is, 10 gm. of chips soaked for 24 hours in 3,000 c. c. of water yielded 32.1 per cent more extract than 10 gm. soaked for the same period in 250 c. c. of water.

It now remains to be determined whether or not experimental results obtained by using quassia extracts on insects will support the preceding data. Preliminary experiments soon proved that quassia extracts are efficient on aphids only; therefore the results dealing with the effectiveness of these extracts on other insects are briefly discussed under the heading, "Pharmacological effects of quassia."

In the various experiments in which many experimental formulas containing quassia extracts were used for the purpose of eliminating all those formulas found to be inefficient in the laboratory, the following aphids were employed: Tulip-tree aphids (*Macrosiphum liriodendri* Mon.), rose aphids (*Macrosiphum rosae* L.), nasturtium aphids (*Aphis rumicis* L.), cabbage aphids (*Aphis brassicae* L.) on kale, pea aphids (*Macrosiphum pisi* L.), aphids (*Aphis* sp.) on bladder senna (*Colutea arborescens* L.), woolly beech aphids (*Phyllaphis fagi* L.), and those aphids (*Chaitophorus populicola* Thos.) found on Carolina poplars. The following leaves, branches, and entire plants, each bearing many aphids, were collected between 7 and 8 a. m. and were placed in bottles of water on a long table by windows: Leaves of tulip trees (*Liriodendron tulipifera*), nasturtiums (*Tropaeolum* spp.), and kale (*Brassica oleracea viridis*); branches of rose-bushes (*Rosa* spp.), bladder senna, beech trees (*Fagus americana*), and Carolina poplars (*Populus deltoides*); and entire sweet-pea plants (*Lathyrus odoratus*) in small pots. The aphids were then sprayed with an atomizer, and the bottles and pots, with their contents, were so arranged on the table that each of them received an equal share of light. The insects were counted before any of them died, and at regular intervals throughout the day those remaining alive were recorded. The three following interfering factors were usually present: (1) To a limited degree the aphids left the leaves and branches and crawled toward the windows; (2) most of the spray solutions were slightly repellent and certainly the less effective ones caused a small percentage of the aphids to migrate from the leaves and branches; (3) the tulip-tree leaves late in the afternoon showed evidence of drying, which consequently caused the remaining live insects to leave them sooner or later. The first factor is insignificant when comparing various results obtained in the laboratory; but, when these results are compared with those obtained outside the laboratory, it must be taken into account. Inside and outside the laboratory the second factor probably has the same weight; but, when the mortality of aphids sprayed is compared with that of those not sprayed, a small probable error should usually be allowed. To overcome most of the error caused by the third

factor, all of the results recorded later than 9 hours after applying the spray solutions were eliminated.

Since the extract of quassia wood (see p. 510) contains constituents other than the supposedly active one called "quassiin," it was considered expedient to begin the experimentation with quassiin in as pure a form as could be obtained on the market. Various solvents were used not only on a small quantity of commercial quassiin powder labeled "purified powder" but also on quassia chips and quassia powder.

(a) EXPERIMENTS WITH QUASSIIN POWDER

Before a discussion of the results obtained by using the extracts dissolved from the quassiin powder it was first thought advisable to determine the solubility of this particular powder in different solvents. According to Schmidt (39), quassiin is difficultly soluble in water or ether, but is readily soluble in alcohol, chloroform, or acetic acid. It is also dissolved by caustic alkalis and concentrated acids, but not by alkaline carbonates.

To determine the solubility of the above-mentioned quassiin in (1) distilled water, (2) in a 0.05 per cent sodium-carbonate solution, (3) in a 0.05 per cent lye solution, and (4) in a soap solution (1.8 gm. of potash-fishoil soap to 1,000 c. c. of water, or 1.6 pounds of soap to 100 gallons of water), the following method was used: An excess of the quassiin powder was placed in a flat 1,000 c. c. bottle containing 500 c. c. of distilled water, and then the bottle with its contents was shaken by means of a mechanical device for 5 hours; this process was repeated three times by using the three other enumerated solvents, one at a time. After filtering the mixture, 100 c. c. of each filtrate obtained were evaporated in a tared dish, and the quantity of the solids remaining in the dish was determined. These solids represented the extract from the quassiin powder plus the solid matter contained in each solvent. It was therefore necessary to determine the quantity of solid matter in 100 c. c. of each solvent not containing quassiin extract, in order to ascertain the weight of the extract from the quassiin powder. The results obtained are summarized in Table V.

TABLE V.—*Solubility of quassiin powder in various solvents*

Solvent.	Solid matter in—		Quantity of extract in 100 c. c. of filtrate.	Solubility of quassiin powder in solvent.
	100 c. c. of filtrate.	100 c. c. of solvent.		
	Gm.	Gm.	Gm.	Parts.
Distilled water.....	0.0322	0.0001	0.0321	1 to 3,084
0.05 per cent sodium carbonate solution.	.1592	.0501	.1091	1 to 917
Soap solution (1.8 gm. to 1,000 c. c.).....	.2735	.1528	.1207	1 to 828
0.05 per cent lye solution.....	.2325	.0501	.1824	1 to 548

According to Table V the addition of an alkali to the water greatly increases the solubility of the quassiin powder—for example, the addition of the lye increased the solubility from five to six times, while the addition of the soap increased it almost four times. It now remains to be determined which one of the four solvents used in dissolving the quassiin powder is preferable in the preparation of spray solutions.

(1) ETHER AND WATER AS SOLVENTS

Owing to the possibility that this quassiin powder may be impure, the procedure detailed below was employed to test its purity. Schmidt (39) says that quassiin is difficultly soluble in ether or water. Merck (24) succeeded in separating a tasteless and supposedly inert substance, which he called "quassol," from impure quassiin powder by using ether.

EXPERIMENT 1.—In an attempt to purify this supposedly impure quassiin powder, 5 gm. of it were extracted with 500 c. c. of ordinary ether for 24 hours, during 6 of which the material was constantly shaken. After filtering, the amount of extract contained in 100 c. c. of the filtrate was determined, and it was found that 1.8 gm., or 35 per cent, of the quassiin powder went into solution in the ether. The ether-soluble residue resulting was of a resinous character and not as bitter as the quassiin powder. Since quassiin is somewhat soluble in ether, it would seem that this residue contained both quassol and quassiin, but probably a greater percentage of the former, because it was not so bitter as the quassiin powder. A small portion (0.57 gm.) of the resinous material was extracted with 500 c. c. of water for 24 hours, during 8 of which the mixture was constantly agitated. After filtering, the filtrate was sprayed upon nasturtium aphids, 58 per cent of which afterwards died (see Table VI). Since this liquid was not very bitter and as it killed only about one-half of the aphids tested, it may be inferred that the extract contained in it was mostly quassol. Since ordinary ether contains a trace of water, it was thought advisable to repeat this experiment by using anhydrous ether for exhausting the quassiin powder. This ether extracted 45 per cent of the powder. A water extract of the ether-soluble portion killed 50 per cent of the nasturtium aphids and 54 per cent of the pea aphids.

EXPERIMENT 2.—In this experiment an extract was prepared from the portion of the quassiin powder not dissolved by the ordinary ether, described in experiment 1, by extracting an excess of it with water for 24 hours, during 8 of which it was constantly shaken. After filtering the mixture, the filtrate, which was much more bitter than that used in the preceding experiment, was sprayed upon nasturtium aphids, all of which afterwards died. An extract was also prepared from the portion of the powder not dissolved by the anhydrous ether, described in the preceding experiment; this killed 95 per cent of the nasturtium aphids and 82 per cent of the pea aphids. Since these residues did not contain quassol, it may be regarded that the extracts used were practically pure quassiin.

(2) WATER AS A SOLVENT

EXPERIMENT 3.—One-tenth gm. of quassiin powder was macerated in 500 c.c. of water for 2 hours with frequent agitation and the mixture was then filtered. Only the undiluted filtrate and one dilution (1:5) of it killed practically all of the aphids tested (see Table VI).

EXPERIMENTS 4 TO 12.—The procedure of these was similar to the one just above, and the results obtained are tabulated in Table VI.

TABLE VI.—Methods of preparing spray solutions from quassia powder with various solvents and results of preliminary tests of these solutions on aphids in laboratory

Experiment No.	Procedure.	Ether solvents, dilutions, and time of extractions.	Species of aphids.	Number of aphids used.	Results.	
					Per-centage killed.	Minimum time required to kill per-centage indicated.
						Hours.
1	Excess of powder shaken in ether, evaporated, and residue extracted with water.	(a) Ordinary ether	<i>Aphis rumicis</i>	79	58	8
		(b) Anhydrous ether.do.....	104	50	7
		(c) Anhydrous ether.	<i>Macrosiphum pisi</i>	62	54	7
2	Powder exhausted with ether and undissolved portions then extracted with water.	(a) Ordinary ether	<i>Aphis rumicis</i>	105	100	8
		(b) Anhydrous ether.do.....	129	95	7
		(c) Anhydrous ether.	<i>Macrosiphum pisi</i>	39	82	7
3	0.1 gm. extracted with 500 c. c. of water for 2 hours and diluted with water.	(a) No dilution....	<i>Macrosiphum liri-odendri</i>	29	100	4
		(b) No dilution....do.....	508	90	8
		(c) No dilution....	<i>Chaitophorus pop-ulicola</i>	335	100	8
4	0.1 gm. extracted with 1,000 c. c. of water for following periods:	(d) Dilution 1:5....	<i>Macrosiphum liri-odendri</i>	39	100	9
		(a) 1 hour.....do.....	16	100	6
		(b) 3 hours.....do.....	59	100	5
5	0.1 gm. boiled in 1,000 c. c. of water for 2 hours and diluted with water.	(c) 5 hours.....do.....	39	100	7
		(d) 24 hours.....do.....	16	100	5
		(a) No dilution....do.....	36	100	2½
6	An excess of powder shaken in distilled water for 5 hours.	(b) Dilution 1:5....do.....	31	100	3
		(c) No dilution....do.....	114	33	8
		(a) 1 hour.....	<i>Aphis</i> sp.	277	82	8
7	0.1 gm. extracted with 1,000 c. c. of soap solution for following periods:	(b) 3 hours.....	<i>Macrosiphum liri-odendri</i>	300	95	8
		(c) 5 hours.....do.....	46	100	3
		(d) 24 hours.....do.....	47	100	2½
8	0.1 gm. boiled in 1,000 c. c. of soap solution for 2 hours and diluted with soap solution.	(c) 5 hours.....do.....	46	100	4
		(d) 24 hours.....do.....	21	100	4
		(a) No dilution....do.....	294	100	2
9	0.1 gm. extracted with 1,000 c. c. of 1 per cent sodium-carbonate solution for following periods:	(b) Dilution 1:5....do.....	111	100	2
		(c) Dilution 1:10....do.....	61	100	2
		(d) Dilution 1:50....do.....	29	100	2
10	0.1 gm. boiled in 1,000 c. c. of 1 per cent sodium-carbonate solution for 2 hours and diluted with water.	(e) Dilution 1:100....do.....	30	100	2½
		(a) 1 hour.....do.....	31	100	4
		(b) 3 hours.....do.....	40	100	4
11	0.1 gm. extracted with 1,000 c. c. of 1 per cent lye solution for following periods:	(c) 5 hours.....do.....	37	100	5
		(d) 24 hours.....do.....	22	100	5
		(a) No dilution....do.....	70	100	2½
12	0.1 gm. boiled in 1,000 c. c. of 1 per cent lye solution for 2 hours and diluted with water.	(b) Dilution 1:5....do.....	29	100	6
		(a) 1 hour.....do.....	54	100	3
		(b) 3 hours.....do.....	128	100	3
		(c) 5 hours.....do.....	101	100	3
		(a) No dilution....do.....	29	100	9

Reference to Table VI shows the following: The ether-water extract (probably mostly quassol) of impure quassia powder killed about one-half of the aphids tested (experiment 1), while the practically pure quassia killed 92 per cent of those sprayed (experiment 2). Generally speaking, all of the remaining extracts were efficient within a few hours. In all but one case it is indicated that boiling slightly increases the efficiency of the extracts. Had they been boiled longer than two hours, this indication might have been reversed. Boiling quassia powder

with lye solution seems to destroy the insecticidal value of the extract obtained, lye probably causing decomposition of the quassia. There seems to be no difference in effectiveness between the unboiled soap-solution and the lye-solution extracts; but the soap-solution extract obtained by boiling the powder for two hours appears to be the most effective of the extracts obtained from this powder. It is thus seen that the addition of lye and soap to the water materially increases the effectiveness of the extracts obtained, while the addition of sodium carbonate to the water only slightly increases the effectiveness of the extract obtained. These results agree with those of the quantitative determinations recorded in Table V, with the exception that the effectiveness of the extract obtained by the sodium-carbonate solution does not correspond to the quantity of extract removed by this solvent.

(b) EXPERIMENTS WITH QUASSIA CHIPS

According to the results of the quantitative determinations (Table III), it was found that the greater the comminution of the quassia material the larger is the quantity of extract capable of being removed. Hence, extracts obtained from quassia powder should be more efficient than those from quassia chips. In the preliminary experiments with aphids, it was ascertained that the former extracts were little, if any, more efficient than the latter extracts. This may be due to two reasons: (1) Since these experiments were performed with such a small number of aphids, the difference in efficiency was not noticeable; and (2) the powder and chips were not from the same identical tree, and probably not from trees of the same species. Since quassia powder will in all probability never be used to any great extent in practical spraying, owing to the expense involved in pulverizing the chips, it will be omitted from the following discussions, and only those results pertaining to the extracts from quassia chips will be briefly described. In all of the following experiments only tulip-tree aphids were used, and for the sake of brevity only the first experiment is briefly stated, and all of them are then summarized in Table VII.

(1) WATER AS A SOLVENT

EXPERIMENT 13.—Twenty-five gm. of chips were macerated in 350 c. c. of water for one-half hour; this process was repeated four times in 1, 3, 10, and 48 hour periods. With the extracts thus obtained the length of time required to kill the aphids tested varied from four to eight hours (see Table VII).

Reference to Table VII shows that the summary of the results of these experiments agrees closely with that of the results of the experiments recorded in Table VI, which deals with quassia. Briefly stated, the similarity of these two summaries is as follows: The addition of lye and soap to the water greatly increases the effectiveness of the extracts obtained, while the effectiveness of the extract dissolved by the sodium-carbonate solution is only slightly better than that of the water extract.

TABLE VII.—Methods of preparing spray solutions from quassia chips with various solvents and results of preliminary tests of these solutions on tulip-tree aphids (*Macrosiphum liriodendri* Mon.) in laboratory

Experiment No.	Object of methods.	Procedure.	Length of time of extractions, dilutions, number of extractions, and strength of solvents.	Number of aphids used.	Length of time required to kill aphids.
					Hours.
13	Effect of length of time of maceration of chips.	25 gm. of chips macerated in 350 c. c. of water for:	(a) $\frac{1}{2}$ hour..... (b) 1 hour..... (c) 3 hours..... (d) 10 hours..... (e) 48 hours.....	283 92 164 128 32	8 7 7 7 4
14	Effect of dilution of water containing extract of chips.	25 gm. of chips macerated in 350 c. c. of water for 24 hours and diluted with water.	(a) Dilution 1 : $\frac{1}{4}$ (b) Dilution 1 : $\frac{1}{2}$ (c) Dilution 1 : 1.....	143 89 67	7 9 9
15	Effect of repeated maceration of chips.	25 gm. of chips extracted with 350 c. c. of water for 2 hours; filtered and extraction repeated.	(a) First extraction..... (b) Second extraction.....	34 37	6 7
16	Effect of boiling and length of time of boiling chips.	25 gm. of chips boiled in 350 c. c. of water; evaporated water replaced.	(a) Boiled $\frac{1}{2}$ hour..... (b) Boiled 1 hour..... (c) Boiled 2 hours..... (d) Boiled 5 hours.....	223 204 77 173	4 4 4 4
17	Effect of diluting boiled water containing extract of chips.	25 gm. of chips boiled in 350 c. c. of water for 2 hours under reflux condenser and then diluted with water.	(a) Dilution 1 : $\frac{1}{4}$ (b) Dilution 1 : 1..... (c) Dilution 1 : 2..... (d) Dilution 1 : 4.....	131 86 142 161	4 4 4 6
18	Effect of repeated boiling of chips.	25 gm. of chips boiled in 350 c. c. of water for 2 hours; filtered and process repeated.	(a) First extraction..... (b) Second extraction.....	339 220	4 4
19	Effect of maceration of chips in soap solution and subsequent boiling.	25 gm. of chips macerated for 24 hours in 350 c. c. of soap solution and boiled for 2 hours.		48	4
20	Effect of diluting soap solution containing extract of chips with water.	25 gm. of chips macerated in 350 c. c. of soap solution for 24 hours and diluted with water.	(a) No dilution..... (b) Dilution 1 : 2..... (c) Dilution 1 : 5..... (d) Dilution 1 : 25.....	101 43 72 36	2 3 5 6
21	Effect of diluting soap solution containing extract of chips with soap solution.	25 gm. of chips macerated in 350 c. c. of soap solution for 24 hours and diluted with soap solution.	(a) Dilution 1 : 2..... (b) Dilution 1 : 5..... (c) Dilution 1 : 10..... (d) Dilution 1 : 25.....	23 18 14 76	2 3 3 5
22	Effect of macerating chips in 1 per cent sodium-carbonate solution with subsequent boiling.	25 gm. of chips macerated in 350 c. c. 1 per cent sodium-carbonate solution for 24 hours and then boiled for 2 hours.		31	5
23	Effect of macerating chips in sodium-carbonate solutions.	25 gm. of chips macerated for 24 hours in 350 c. c. of sodium-carbonate solution of the following strength:	(a) 1.0 per cent..... (b) 0.5 per cent..... (c) 0.3 per cent..... (d) 0.1 per cent..... (e) 0.05 per cent.....	102 23 64 15 22	4 5 6 5 5
24	Effect of macerating chips in sodium-carbonate solution and then diluting with water.	25 gm. of chips macerated in 350 c. c. of 0.05 per cent sodium-carbonate solution for 24 hours and diluted with water.	(a) No dilution..... (b) Dilution 1 : 5..... (c) Dilution 1 : 25.....	56 53 43	5 6 9
25	Effect of macerating chips in lye solutions	25 gm. of chips macerated for 24 hours in 350 c. c. lye solution of following strength:	(a) 0.5 per cent..... (b) 0.3 per cent..... (c) 0.1 per cent..... (d) 0.05 per cent.....	24 28 17 34	2 2 2 2
26	Effect of macerating chips in lye solution and then diluting with water.	25 gm. of chips macerated in 350 c. c. of 0.05 per cent lye solution for 24 hours and diluted with water.	(a) No dilution..... (b) Dilution 1 : 5..... (c) Dilution 1 : 25.....	81 93 33	2 6 8

With the exception of the effectiveness of the extract removed by the sodium-carbonate solution, these results agree with those of the quantitative determinations recorded in Table V. Furthermore, all of these results closely agree with the statement made by Schmidt (39), who

claims that quassia is slightly soluble in water, and is readily soluble in the caustic alkalies, but is not soluble in the alkali carbonates. Table VII further shows that extracts from chips boiled for 5 hours are slightly more effective than those from chips soaked for 24 hours, results agreeing with those of the quantitative determinations. But the effectiveness of the first extract from chips soaked for 2 hours is only slightly better than that of the second extract, whereas the ratio should be 4 to 1, in order to agree with the quantitative determinations; however, experiments performed in the laboratory on a large scale (p. 515) with the first and second extracts give a ratio of about 5 to 2. The most important result recorded in Table VII is that the soap-solution extract and lye-solution extract are equally effective, but, when the solutions containing the extracts are diluted, the former with soap solution and the latter with water, it is readily seen that the dilutions containing the soap-solution extract are much more effective and more economical, because they already contain the necessary "spreader," while the lye dilutions to be equally effective must have soap added to them before they are applied.

3.—EFFECTIVENESS OF SOME ECONOMIC FORMULAS

The preceding preliminary experiments clearly show that extracts from quassia chips soaked in water are less effective than those from chips boiled in water; but, on the other hand, extracts from chips soaked in soap solution, sodium-carbonate solution, and lye solution are more effective than those from chips boiled in these three solvents. This seems to indicate that at a high temperature alkalies decompose quassia. As already stated, soap-solution and lye-solution extracts, not boiled, are the most effective ones found; and of these two extracts the former is the more economical and perhaps the more efficient for practical work. The soap-solution extract was further tested in the laboratory before it was applied in practical work, but the lye-solution extract did not seem to warrant further tests.

The following experiments were therefore performed with variously concocted formulas containing soap-solution extract to determine whether or not quassia extract may be employed as a general insecticide for all aphids. In each formula the soap was used in the proportion of 1.6 pounds to 100 gallons of water. This amount of soap has no detrimental effect upon the plants sprayed and very little upon the aphids.

To be able to compare more accurately the effectiveness of the various formulas, experiments were first performed in the laboratory on a small scale, and then outside the laboratory on a larger scale; with this method the live insects in the laboratory were counted at regular intervals, but outside the laboratory they were generally estimated. In order to have a standard by which the efficiency of quassia extract might be judged, nicotine sulphate in soap solution and also in water was sprayed upon aphids.

(a) EFFECTIVENESS OF SPRAY SOLUTIONS APPLIED IN LABORATORY

The aphids were collected and were sprayed as described on page 508. The number used for each individual test varied from 124 to 436, with 253 as an average. Reference to Table VIII shows the following: Of the four species sprayed, the mortality of *Macrosiphum liriodendri* was the lowest and that of *Aphis* sp. the highest; the "wool" on *Phyllaphis fagi* seemed to prevent the spray solution from thoroughly wetting these aphids. From the laboratory viewpoint formulas 1A and 3A (first extracts) were efficient, but only upon two of the four species sprayed. For each of the four formulas the solution containing the second extract was less effective than that containing the first extract, indicating that more of the toxic principle was removed from the chips during the first than the second extraction. Formula 3A (first extract), the one used by Parker (32) on the hop aphid, was efficient upon only *Aphis* sp. and *Chaitophorus populicola* in the laboratory, but it is shown on page 517 that this formula is not efficient upon the same species of *Aphis* outside the laboratory and probably not upon *C. populicola*, although the latter species was not sprayed outside the laboratory.

TABLE VIII.—Effectiveness of quassia extracts, soap solution, and nicotine sulphate applied in the laboratory

Formula No. and extract No.	Quantity of quassia chips used.	Quantity of fish-oil-soap solution (1.6 pounds of soap to 100 gallons of water) used as solvent.	Length of time chips soaked.	Stock solution.	Final dilution with fish-oil-soap solution.	Quantity of chips used to 100 gallons of water.	Percentage of aphids dead 9 hours after application of spray solutions.			
							<i>Macrosiphum liriodendri</i> on tulip-tree leaves.	<i>Aphis</i> sp. on bladder-senna branch.	<i>Phyllaphis fagi</i> on beech-tree leaves.	<i>Chaitophorus populicola</i> on Carolina poplar branches.
1A (first extract)...	Gm. 25.0	C. c. 350	Hours. 24	C. c. 350	Parts. 1 : 50	Pounds. 1.26	74	100	76	98
1B (second extract).....	25.0	350	24	350	1 : 50	1.26	56
2A (first extract)...	25.0	350	24	350	1 : 35	1.81	81
2B (second extract).....	25.0	350	24	350	1 : 35	1.81	48
3A (first extract)...	6.3	a 2,000	24	2,000	None.	2.80	90	100	86	99
3B (second extract).....	6.3	2,000	24	2,000	...do...	2.80	76
4A (first extract)...	25.0	350	24	350	1 : 20	3.16	92
4B (second extract).....	25.0	350	24	350	1 : 20	3.16	77
Fish-oil-soap solution (control)....	1	5	3	3
Nicotine sulphate (1 : 1,200 of soap solution).....	95
Nicotine sulphate (1 : 1,200 of water).....	60	100

a In this formula the chips were soaked for 24 hours in 2,000 c. c. of water, and the soap was added subsequently.

(b) EFFECTIVENESS OF SPRAY SOLUTIONS APPLIED OUTSIDE THE LABORATORY

In the experiments described under this heading various plants, all badly infested with aphids, were sprayed early in the morning with a hand sprayer, and the following morning the number of aphids killed was estimated. Reference to Table IX shows the following: Formula 3A, the one used by Parker (32) on the hop aphid, was efficient upon only *Aphis rumicis*, but killed 95 per cent of the aphids on the ragweed and asters. Formula 6B (first extract) was efficient upon the aphids on the ragweed and asters, but killed only 95 per cent of the pea aphids, *Aphis* sp., and rose aphids tested; the second extract of the same chips was not efficient upon any of these aphids. Formula 7 was efficient upon all the aphids tested, but the quantity of chips employed is so great that the formula could not be used economically for practical spraying. Not one of those formulas, even including nicotine sulphate (1:1,200 of soap solution) was efficient upon *Myzus persicae* on the eggplant.

(c) COMPARATIVE RAPIDITY OF ACTION OF QUASSIA EXTRACT AND NICOTINE SULPHATE

Since formula 6B (first extract, Table IX) is somewhat less expensive (excluding the labor of preparing it) than nicotine sulphate (1:800 of soap solution), and, as its efficiency 24 hours after its application is comparable to that of the nicotine-sulphate solution, the following experiments were performed in the laboratory to determine the rapidity of the action of these two insecticides, so that the effects of a shower upon aphids sprayed with these two solutions might be deduced. In each individual experiment the aphids were first sprayed with one or the other insecticide and then with tap water after one of the intervals of 5, 10, 20, 30, 60, or 120 minutes. The following plants and aphids were used: Kale leaves bearing 180 to 425 aphids; rose branches bearing 22 to 115 aphids; nasturtium leaves bearing 89 to 107 aphids; and bladder-senna branches bearing 36 to 92 aphids.

Twenty-four hours after applying formula 6B, the following aphids were dead: None of those sprayed with tap water after 5, 10, and 20 minute intervals; a few nasturtium aphids sprayed with tap water after a 30-minute interval; a few of those on the kale leaf and bladder-senna branch and about one-half of those on the nasturtium leaf sprayed with tap water after a 60-minute interval; most of those on the kale leaf and bladder-senna branch, practically all on the nasturtium leaf, but only a few of the rose aphids sprayed with tap water after an interval of 120 minutes. All the aphids on a kale leaf sprayed with this formula, but not later with tap water, were dead 6 hours after being sprayed; all of those on another leaf sprayed with the nicotine-sulphate solution, but not later with tap water, were dead 2 hours after being sprayed.

TABLE IX.—Effectiveness of quassia extracts, soap solution, and nicotine sulphate applied outside the laboratory

Formula No. and extract No.	Quantity of quassia chips used.		Length of time chips soaked.	Quantity of fishoil-soap solution (1.6 pounds of soap to 100 gallons of water) used as solvent.		Stock solution.	Final dilution with fishoil-soap solution.	Quantity of chips used to 100 gallons of water.	Approximate number of aphids sprayed and percentage of aphids dead 24 hours after application of spray solutions.															
	Gm.	C. c.		HOURS.	C. c.				Parts.	Lbs.	<i>Aphis rumicis</i> on dwarf nasturtiums.		<i>Macrosiphum ambrosiae</i> on giant ragweed.		<i>Macrosiphum</i> sp. on aster plants.		<i>Macrosiphum pisi</i> on sweet peas.		<i>Aphis</i> sp. on bladder senna.		<i>Macrosiphum rosae</i> on rose-bushes.		<i>Myzus persicae</i> on eggplant in greenhouse.	
											Number sprayed.	Percentage killed.	Number sprayed.	Percentage killed.	Number sprayed.	Percentage killed.	Number sprayed.	Percentage killed.	Number sprayed.	Percentage killed.	Number sprayed.	Percentage killed.	Number sprayed.	Percentage killed.
1A (first extract).....	25.0	350	24	350	1 : 50	1.26	Thousands.	25	Hundreds.	70	160	25	780	20	Hundreds.	25				
2A (first extract).....	25.0	350	24	350	1 : 35	1.81	160	50				
3A (first extract).....	6.3	2,000	24	2,000	None.	2.80	Thousands.	99	Hundreds.	95	300	95	320	90	160	75	180	40	Hundreds.	25				
4A (first extract).....	25.0	350	24	350	1 : 20	3.16	160	90	2,700	50				
5 (first extract).....	25.0	350	24	2,000	None.	11.00	1,340	80	1,540	80				
6A (first extract).....	50.0	350	24	2,000	None.	22.00	1,120	90	1,500	90				
6B (first extract).....	50.0	2,000	24	2,000	None.	22.00	Hundreds.	99	300	100	320	95	1,120	95	1,500	95	Hundreds.	25				
6B (second extract).....	50.0	2,000	24	2,000	None.	22.00	do.	90	300	40	1,120	70	1,500	90				
6C (first extract).....	50.0	350	6	2,000	None.	22.00	1,120	90	1,500	90				
6D (first extract).....	50.0	2,000	6	2,000	None.	22.00	1,120	95	1,500	94				
7 (first extract).....	25.0	350	24	350	None.	63.30	Thousands.	100	300	100	1,120	95	180	100				
Fishoil-soap solution (control).	do.	5	160	5	Hundreds.	0				
Nicotine sulphate (1 : 1,200 of soap solution).	160	95	do.	35				
Nicotine sulphate (1 : 800 of soap solution).	1,900	98	1,350	95				
Nicotine sulphate (1 : 1,200 of water).	1,120	60				

^a In this formula the chips were soaked for 24 hours in 2,000 c. c. of water, and the soap was subsequently added.

Twenty-four hours after applying the nicotine-sulphate solution the following aphids were dead: None of those sprayed with tap water after 5 and 10 minute intervals; a few of those on the kale and nasturtium leaves sprayed with tap water after a 20-minute interval; several of those on the kale leaf and nearly all on the nasturtium leaf sprayed with tap water after a 30-minute interval; nearly all belonging to the four species sprayed with tap water after a 60-minute interval; and all belonging to the four species sprayed with tap water after a 120-minute interval.

The foregoing results show that nicotine sulphate acts very quickly, while quassia extract acts very slowly. These results also indicate that a shower 2 hours after the application of these insecticides does not affect the efficiency of the nicotine sulphate, while it greatly reduces the efficiency of quassia extract.

PHARMACOLOGICAL EFFECTS OF QUASSIIN

The preceding experiments show that quassia extract kills aphids only by coming in contact with them, but it still remains to be shown how it kills them. This phase of the work involves a careful study of the physiological effects of quassiin on aphids and of what tissue is vitally affected.

I.—PHYSIOLOGICAL EFFECTS OF QUASSIIN

To determine how quassiin in the form of powder and in spray solutions affects insects when dusted or sprayed upon them, the physiological effects of this substance on the insects were observed. Of the various insects used in the experiments it was ascertained that quassiin is fatal only to aphids; consequently the following discussion of results will be devoted chiefly to this family of insects, and the effects of this substance on the other insects utilized will be noted only here and there.

(a) EFFECTS OF QUASSIIN POWDER

At the outset a purified powder of quassia, already mentioned on page 509, was used. This powder is light yellow and is supposed to be largely quassiin. It is intensely bitter, has a faint odor, and is disagreeable to work with, for after a few moments, regardless of how carefully it is handled, a bitter taste is experienced which sometimes lasts half a day, and consequently a headache often results. It would thus seem that the odor, and probably the minutest particles of the powder suspended in the air pass into the nose and mouth and give rise to the sensation of having tasted it.

To determine whether the exhalation from the quassiin powder alone is sufficient to kill aphids, a watch glass was completely filled with the powder; then a wire screen was laid over the powder and a nasturtium leaf, bearing about 45 aphids (*Aphis rumicis*), was laid upon the screen

so that the aphids were against the wire screen but not in contact with the powder. Fifty minutes later most of the aphids were "stupid"; 5 hours later 2 of them were dead; and the following morning 15 were dead, and the others had crawled away.

At 11 a. m. leaves from nasturtiums, apple, ragweed, and snowball, all bearing many aphids (*Aphis rumicis*, *Aphis pomi* De Geer, *Macrosiphum ambrosiae*, and *Aphis viburniphila* Patch), were dusted with the quassia powder. Forty-five minutes later most of the aphids were inactive, and by 4.30 p. m. practically all of them were dead.

At 12 noon leaves from mulberry and pear trees, bearing many fall webworms (caterpillars of *Hyphantria cunea* Dru.) were dusted with the quassia powder. By 2.30 p. m. nearly all of the webworms were inactive, and by 4.30 p. m. all of them were apparently dead; the following morning at 10 o'clock all were still apparently lifeless, except a few which were slowly reviving from their "stupor"; two days later still most of them had revived and were as active as usual.

At 4 p. m. several mulberry leaves were cut into small pieces and then some of the quassia powder was mixed with them; next, 20 medium-sized silkworms (larvæ of *Bombyx mori* L.) were placed upon the mixture of leaves and powder. Half an hour later three silkworms were apparently lifeless, and the others appeared weak and sick; they did not seem to eat the bits of leaves, but crawled away from them, and consequently it was necessary to confine them in a box with the poisoned food. The following morning all of them were apparently dead, and they never revived thereafter.

A microscopical examination shows that the minutest particles of the quassia powder are sufficiently small to pass through the spiracles of aphids and into the tracheal trunks for some distance; but, before they can come into contact with the nervous system (supposing that this system is the tissue vitally affected), they must pass through the smaller tracheal branches and even then through the tracheal walls in order to come into contact with the nerve cells. Furthermore, it is scarcely possible that they pass even through the spiracles of aphids and fall webworms because the spiracles are well guarded by hairs (22, p. 104). In view of the preceding, it seems that the exhalations alone from this powder killed the aphids and silkworms employed and rendered the fall webworms inactive for about a day.

(b) EFFECTS OF QUASSIA POWDER

The preceding experiments were repeated by dusting quassia powder upon aphids from nasturtiums and snowballs, and upon silkworms and fall webworms. During the first 24 hours no effects were observed, except that a few of the smallest aphids died and that the silkworms were rendered slightly "stupid" for a few hours, but the webworms apparently were not affected at all. Some of the aphids were even

buried in the powder; after a few moments they invariably came to the surface and crawled away from the powder without apparently being affected.

This powder is made from quassia chips finely ground. It has a very faint odor, but while handling it a person does not experience a bitter taste as he does while working with quassiin powder. The minutest particles of the powder are no larger than those of the quassiin powder, but the largest particles are considerably larger than those of the quassiin powder. While the particles of the quassiin powder adhere to one another considerably, those of the quassia powder do not.

(c) EFFECTS OF WATER EXTRACT OF QUASSIIN POWDER

A strong extract was secured by boiling for two hours 200 c. c. of water containing 1 gm. of the quassiin powder. When the mixture was filtered, the filtrate was not quite as clear as water; it emitted a very faint odor and had a bitter taste. When applied with an atomizer it seems that some of the fine spray carried by the air must have passed into the operator's mouth, because a bitter taste was always experienced whenever this solution was used as a spray.

To determine whether the exhalations or vapor from the preceding solution is alone sufficient to kill aphids, a 75-c. c. beaker was filled with this spray mixture. A wire screen was then laid over the liquid and a nasturtium leaf bearing many aphids was placed upon the wire screen so that the insects were near the liquid, but not in contact with it. Not one of the aphids apparently was affected.

To ascertain whether the quassiin contained in the preceding solution is volatile when heated, 50 c. c. of the solution were poured into a 100-c. c. retort and heated. It was found that the steam produced by heating the solution had no apparent effect upon the aphids (*Aphis rumicis* and *Macrosiphum liriodendri*) tested. The steam was odorless, and, when condensed, the resultant liquid was as clear as water. It was tasteless and lead acetate did not precipitate it, while the same compound slightly precipitated the quassiin solution. It is thus seen that the quassiin contained in the solution is nonvolatile, and therefore such solutions can not be used for fumigating purposes. Furthermore, it follows that the vapors arising from quassia extract solutions do not carry quassiin.

At 9 a. m. 508 aphids upon tulip-tree leaves were sprayed with quassiin solution. At 12 noon only a few of the aphids were dead, at 4 p. m. 90 per cent were dead, and the following morning all were dead. It was noted that a faint odor was emitted by the spray solution upon the leaves, and even when the leaves became dry a very faint odor resembling that of the quassiin solution was still given off.

On account of the long time required for the quassiin to kill the aphids, it is probable that this insecticide acted as a stomach poison. The spray solution might have passed through the stomata and epidermis of the

leaves, and when within the interior of the leaves it might have been imbibed through the proboscides of the aphids and then passed into their alimentary canals. To test this probability, 58 aphids on tulip-tree leaves were gently brushed into a wire-screen cage, and at 8.30 a. m. they were sprayed with the quassiin solution; at 12.30 p. m. only a few of them were dead, but at 4.30 p. m. 88 per cent of them were dead. This experiment shows that the quassiin did not act as a stomach poison by passing from the interior of the leaves through the proboscides and then into the alimentary canals of the aphids, and it is unlikely that the aphids imbibed some of the solution on their bodies before they died. While tracing insecticides inside aphids, the senior writer has never seen the poisons either in the proboscis or in the alimentary canal. Sections also show that quassia-extract solutions do not pass into the interior of plants.

In the foregoing it is shown that neither the exhalations nor vapor from the quassiin solution kill aphids, and this insecticide when applied as a spray also does not act as a stomach poison; but there yet remain the two following possible ways in which it may exert its effects upon aphids: (1) The solution may enter the spiracles and come into contact with the nervous system or it may cause death by suffocation, or (2) some of the fine spray may be taken into the respiratory system while the aphids are being sprayed.

Regarding the first view, the senior writer (22, p. 103) has recently shown that nicotine-spray solutions (not containing soap) do not enter the spiracles of aphids, and it is shown on page 525 that quassia-extract solutions (not containing soap) also do not enter the spiracles. If it is granted that this solution does gain entrance through the spiracles but does not come into contact with the nerve cells, would it kill aphids within a limited time by cutting off the supply of oxygen? To test this possibility one sweet-pea plant and four nasturtium leaves, each bearing many aphids, were submerged in water for $5\frac{1}{2}$ hours. A film of air surrounded the leaves, stems, and parts of the aphids, but it was gradually absorbed by the water, so that at the time at which the insects were removed from the water practically all of the air had been absorbed, except several minute bubbles which still adhered to the aphids. When removed from the water, one-half of the pea aphids had fallen from the plant to the bottom of the jar containing the water; they were apparently lifeless, while those remaining on the plant moved when touched. All of the nasturtium aphids exhibited signs of life when removed from the water. Within a short time the inactive aphids revived, and all of the submerged ones soon became normal. It is therefore evident that aphids are not easily suffocated.

To support the second view, three nasturtium leaves and two sweet-pea plants, each bearing many aphids, were submerged in the quassiin solution for half a minute. The solution did not adhere well to the leaves and

aphids, and the insects afterwards were apparently not affected. For this solution to be effective it must be sprayed upon the insects, and since it can not reach the nervous system in any form other than as a fine spray carried by the air, it is believed that death occurred as a result of some of the fine spray being taken into the respiratory system while the solution was being applied. This view agrees with the one that some of the fine spray which is carried by the air passes into the operator's mouth, thereby causing a bitter taste.

At this place should be mentioned quassiin extract as a stomach poison for bees and flies. A large quantity of quassiin, extracted from the quassiin powder and then dissolved in sugar sirup, was fed to 250 honeybees (*Apis mellifica* L.) in observation cases. At no time was a symptom observed which could be attributed to the effects of the quassiin, and these bees lived practically as long as controls fed sugar sirup not containing this insecticide. These results agree with those obtained by Illingworth (18, p. 160), who fed sweetened quassia-extract solution to flies (*Rhagoletis pomonella*). He says:

The flies ate the sweetened, bitter liquid freely, but no harm came to them.

It should be noted, however, that Brande (3) in an early textbook states that quassia is an effectual stomach poison for flies.

(d) EFFECTS OF WATER EXTRACT OF QUASSIA CHIPS

Fifty grams of quassia chips were soaked in 1,000 c. c. of water for 24 hours; this proportion is about equal to 44 pounds of chips to 100 gallons of water. At 8 o'clock many rose aphids, fall webworms, and larvæ of the potato beetle (*Leptinotarsa decemlineata* Say) were sprayed with the above solution; at 11 o'clock few of the aphids were apparently dead and many of the webworms were inactive; at 1 o'clock all of the aphids and webworms were apparently dead, but only a few of the former had fallen from the plants; all of the potato-beetle larvæ had fallen to the ground, some were apparently dead and the others were so affected that they could yet move their legs but could not crawl; at 4.30 o'clock all of the insects sprayed were apparently dead. The following morning several of the aphids and practically all of the potato-beetle larvæ had revived, while the webworms were slowly recovering from the effects of the spray. On the second day after being sprayed, most of the webworms had become normal.

(e) EFFECTS OF SOAP-SOLUTION EXTRACT OF QUASSIA CHIPS

In the preceding paragraph and on pages 512 to 513 it is shown that strong water extracts of quassia chips do kill aphids when properly applied, but it is also shown on pages 514 to 515 that weaker extracts containing soap solution are really more effective. As a rule, while the inefficiency of quassiin dissolved in water may be attributed to the poor

insecticidal properties of this substance and to the fact that water does not spread it well, the efficiency of it when dissolved in soap solution would seem to be due to the fact that the soap solution spreads it in such a manner that a larger amount of it may reach the nervous system. It now remains to be shown how it reaches the nervous system, for there is no other plausible way of explaining its effects on aphids.

Experiments in which a soap-solution extract of quassia chips was used, in the same manner as described on pages 514 and 515, demonstrate that the exhalations from this spray mixture do not kill aphids, and that the mixture does not act as a stomach poison when applied as a spray. On page 515 it is further shown that soap solution containing no quassia extract but the same amount of soap as employed in the soap-solution extract has no apparent effect on some aphids, and but very little on other aphids.

While the spray mixture was being applied, some of the fine spray might have been taken into the respiratory system and possibly came into contact with the nerve cells, but, comparing the effects of this spray mixture with that containing no soap, the writers are inclined to believe that a larger amount of the quassia extract is required in order to produce the results observed. Owing to the fact that soap solutions have weak surface tensions, they adhere well to the surfaces of plants and insects; they spread readily, and consequently should pass freely into the spiracles of insects. Under the following heading it is shown that they not only pass into most of the larger tracheæ but also come into direct contact with the nerve cells.

The senior writer (22, p. 92) attributed the abnormal behavior exhibited by aphids which had been sprayed with a solution of nicotine to motor paralysis, but at no time while using quassia on aphids was a similar behavior observed. While nicotine acts quickly and causes pronounced symptoms, quassia acts very slowly, and the behavior of aphids poisoned by it is so normal that the few abnormal reactions exhibited are generally overlooked. Soon after being sprayed with a nicotine solution, aphids remove their beaks from the plants and wander about considerably, and sooner or later they fall paralyzed from the plants, after which they soon die. Aphids sprayed with a solution of quassia extract are no more irritated than when they are sprayed with water. While being sprayed, they lie flat on the leaves, and later seldom remove their beaks from the plants; consequently they wander about little. Usually they do not begin falling from the plants till three or four hours after being sprayed, and by that time most of those that fall are dead. As a rule they die quietly with the beaks stuck into the plants, and in most cases it is necessary to touch them before being able to decide whether they are dead or alive. While practically all of the aphids sprayed with a nicotine solution fall from the plants before they die, not

more than one-half of those sprayed with a solution of quassia extract usually fall from the plants until several hours after they are dead.

The preceding symptoms exhibited by aphids do not indicate that the nervous system is the first tissue to be vitally affected, but a critical study of the behavior of pea aphids sprayed with a quassia solution shows one symptom which indicates a nervous affection. Several sweet-pea plants bearing many aphids were sprayed with a solution containing a definite proportion of the extract from quassia powder in water. An hour after being sprayed, a few of the aphids stood up and trembled vigorously for two or three seconds; afterwards they gradually became less active when touched and weaker until they died. The trembling behavior was never observed in unsprayed aphids.

2. HISTOLOGICAL METHODS OF TRACING QUASSIA IN TISSUES

To determine, if possible, what tissue is vitally affected when aphids are sprayed with solutions containing extracts of quassia powder and quassia chips, the insects, after being treated with these solutions, were fixed in a fluid containing a precipitant. By this means one or more constituents in each spray solution were precipitated wherever they had gone into the insects; and after carefully studying the microscopical sections made from these aphids, it was usually easy to trace the precipitated particles.

(a) TRACING WATER EXTRACTS OF QUASSIA POWDER INTO APHIDS

Of the few reagents that precipitate quassia, not one of them precipitates it satisfactorily so that the above procedure might be followed. Owing to the unsatisfactory precipitation, it was necessary to add to the spray solution some chemical which under normal conditions is never used, in order that this chemical might be thrown down wherever the spray solution had carried it into the insect.

The senior writer (22, p. 101, 103) submerged aphids for 45 minutes in, and also sprayed them with, a nicotine solution colored with indigo-carmin. By using the above method it was found that the solution had passed into a few of the larger tracheæ of the aphids submerged, but never into the tracheæ of those aphids sprayed, and only once in the latter had a little precipitate lodged in a spiracle. These results indicate that any solution almost totally water sprayed upon aphids would not pass into the tracheæ, unless the permeability of this solution had been considerably increased, or its surface tension had been rendered very weak by the addition of some other substance not already in the solution.

Despite the supposition that water containing quassia has practically the same permeability and surface tension as does water not containing this substance, it was desirable to know whether this solution would pass into the tracheæ. After failing to obtain a satisfactory precipitate by

mixing either lead acetate or tannic acid with the solution containing the water extract of quassiin powder (described on p. 520), 4 parts of the quassiin solution were added to 1 part of aqueous ferric-chlorid solution. Aphids submerged in this mixture for $\frac{1}{2}$ minute, 30 minutes, and 40 minutes were then fixed in a fluid made in the proportion of 5 c. c. of absolute alcohol to 5 drops of tannic acid. A black precipitate is thrown down when this acid unites with ferric chlorid. After remaining in the mixture of alcohol and tannic acid an hour, the aphids were removed and were then placed into absolute alcohol to insure better fixation. Since this precipitate, as well as the other precipitates, do not adhere well to insects, no liquid was used for straightening the ribbons on the slides; the sections were pressed against the slides by using the fingers, and as alcohol easily removes such precipitates from sections, most of the sections were not stained, and these were left in the xylol only a sufficient time to remove the paraffin. Those sections stained were usually not reliable for tracing precipitates, because some of the precipitate had been lost while the slides were being run through the alcohols, and, furthermore, very often the stain masked the precipitate.

A study of the foregoing sections showed a small amount of black precipitate on the outside of the integuments of the aphids that had been submerged only one-half minute, but none was seen in the spiracles or in the tracheæ; however, in those aphids submerged for 30 and 40 minutes much more black precipitate was seen on the outside of their integuments, and occasionally a few small particles in the spiracles and in the tracheæ a short distance from the spiracles.

(b) TRACING SOAP SOLUTION EXTRACTS OF QUASSIA CHIPS INTO APHIDS

Aphids on sweet peas and nasturtiums were sprayed so heavily with the soap-solution extract (1A) from quassia chips described on page 515 that the solution collected in drops around the legs of the aphids and had not all disappeared $3\frac{1}{2}$ hours later, when the aphids and leaves were fixed. Some of these insects were sprayed with the solution colored with carmine acid; and the others with the solution not containing a stain. The former were fixed in absolute alcohol overnight, and the latter in the fixative described below. Absolute alcohol containing ferric chlorid ($\text{Fe}_2\text{Cl}_6 + 12 \text{H}_2\text{O}$) readily precipitates the potassium in the soap solution; but since the water in the ferric chlorid dilutes the absolute alcohol, a good fixation does not result. To avoid this difficulty the ferric chlorid was melted and fused, thereby dehydrating it. Absolute alcohol was then saturated with the resulting cold melt, and after filtering the mixture an amber colored liquid resulted. When this liquid was mixed with the soap solution, a yellowish, flocculent precipitate was formed. Those aphids fixed in this fluid were afterwards well washed in pure absolute alcohol to remove the fixative and to insure better fixation.

A study of the sections from the above aphids shows that the precipitate did not adhere well to the tissues and, owing to its color, it is discernible in sections only with difficulty. Nevertheless, small particles of it were observed occasionally on the integuments and in the tracheæ, showing that a soap solution not containing a stain passes into the respiratory system of aphids when used as a spray.

Owing to the color and adhering ability of precipitated carmine acid, the sections from the aphids sprayed with the soap-solution extract colored with this stain are much more satisfactory to study. A careful study of them shows the following: Much of the red precipitate adheres to the outside of the integuments, but none of it has passed through them; much of it also lies in the spiracles and at various places in practically all the large tracheæ in the abdomen, thorax, and head, and in the bases of all the legs, but none was observed in the lumens passing through the proboscides. In the abdomens and bases of the legs the fat cells surrounding the tracheæ are generally stained red, indicating that the liquid had passed through the tracheal walls. At one place in a thorax a trachea, bearing some of the precipitate, runs along beside a large muscle which is also slightly stained. More or less precipitate was also observed in the tracheæ lying against the optic lobes, the brain, and thoracic ganglion; and occasionally it was noted that the colored liquid had passed through the tracheal walls and had stained the nerve cells near by. Only a few small particles of the precipitate were observed in the interior of the optic lobes and brain.

The preceding results indicate that the nerve tissue is the one vitally affected, because the spray solution does not seem sufficiently distributed in the other tissues to cause fatality, whereas only a few particles of any toxic substance in the brain and ganglia usually cause death.

SUMMARY

Throughout this investigation the experimental results obtained almost invariably support the results of the quantitative determinations. The data obtained pertaining to this portion of the work are as follows:

(1) Medium-sized quassia chips (samples 3 and 4, Table III) soaked for two hours in water yield during the first extraction about 60 per cent of their total soluble matter, but only about 15 per cent during the second extraction; if soaked for 24 hours, they yield during the first extraction about 60 per cent, but during the second extraction about 10 per cent. Experiments with first and second extracts always showed that the first extract was the more effective, but the ratio of effectiveness of the first to the second extract was only about 9 to 7; this is true for both water extracts and soap-solution extracts. In practical spraying neither one of the water extracts nor the second soap-solution extract was efficient, but the first soap-solution extract was sometimes efficient.

(2) Quassia chips boiled longer than four hours in water yield but little more extract than those boiled for just four hours, and the quantity obtained is about 1.5 times that obtained from chips soaked in water for 24 hours. Extracts from chips soaked in water are usually less effective than those from chips boiled in water, but not one of those tested would be efficient in practical work.

(3) The smaller the quassia chips and the finer the quassia powder used, the greater is the quantity of extract removed.

(4) The larger the volume of water used as a solvent, the greater is the quantity of extract removed; for example, 10 gms. of chips soaked for 24 hours in 3,000 c. c. of water yield 32.1 per cent more extract than 10 gms. soaked for the same period in 250 c. c. of water. The practical experiments well support this view.

(5) The solubility in water of the quassiin powder used in these experiments was found to be 1 to 3,000. In a 0.05 per cent sodium-carbonate solution, a soap solution (1.8 gm. of soap to 1,000 c. c. of water), and in a 0.05 per cent lye solution its solubility was, respectively, 3, 4, and 5 times as great.

The experimental results obtained with quassiin powder and quassia chips supported this view in only a general way. The sodium-carbonate solution extract was only slightly more effective than the water extract. The soap-solution extract and lye-solution extract were equally effective in the laboratory when applied as prepared; but when the former solution was diluted with soap solution and the latter solution with water, the dilutions containing the soap-solution extract were much more effective and also more economical, provided the soap was also added to the dilutions containing the lye-solution extract. Extracts from chips soaked in the solvents mentioned are more effective than those from chips boiled in these solvents. This seems to indicate that at a high temperature alkalis decompose quassiin, or render it insoluble.

The following results deal with the pharmacological effects of quassiin:

(1) A moderately bitter and practically ineffective extract was dissolved from commercially pure quassiin powder, leaving a residue whose water extract was intensely bitter and quite effective. The first extract corresponds to quassol, a supposedly inert and tasteless substance with a slight admixture of quassiin; the second extract corresponds exactly to pure quassiin.

(2) The exhalations alone from the quassiin powder killed aphids, but the exhalations from quassia chips, quassia powder, and those from solutions containing quassiin extract and quassia extract were ineffective. Quassia powder dusted upon insects is ineffective, while quassiin powder is quite effective, indicating that the exhalations pass into the respiratory system and that they then affect the nervous system. The minutest particles of either powder are sufficiently small to pass into the spiracles, but they do not cause death by closing the entrances of the tracheæ.

(3) Quassia and quassiin spray solutions, not containing soap, kill aphids when applied sufficiently strong. By the process of elimination it is concluded that death occurs as a result of some of the fine spray being breathed into the respiratory system while the aphids are being sprayed.

(4) The greater effectiveness of solutions containing soap is due to the weaker surface tension of such solutions, which pass freely through the spiracles and finally reach the nervous tissue, where they kill by slowly affecting the nerve cells.

(5) While nicotine acts quickly and causes pronounced symptoms, quassiin acts very slowly and causes but few symptoms, and these are never pronounced. While nicotine kills by paralysis, quassiin causes no noticeable paralysis, but aphids poisoned by it slowly become inactive and finally die in what is known as "coma" in the higher animals.

In conclusion, it should be stated that owing to the poor insecticidal properties of quassiin, quassia extract can never become a general insecticide for all aphids. Of course, the amount of extract to be used could be sufficiently increased so that the spray solution would perhaps be efficient on any particular aphid, but in most cases the expense would prohibit its use. The most effective formula (6B, first extract, Table IX) used by the writers was prepared by soaking 22 pounds of quassia chips in 100 gallons of fish-oil-soap solution (1.6 pounds of soap to 100 gallons of water) for 24 hours. This spray solution under the most favorable conditions was efficient on only two of the six species of aphids tested, but the results as recorded are comparable to those obtained by using nicotine-sulphate solution. Nevertheless, owing to the slow action of quassiin, this spray solution is much less reliable than is nicotine-sulphate solution, because the aphids sprayed have better opportunities to migrate, and should it rain a few hours after the solution has been applied its effectiveness would be greatly reduced, while such is not true for nicotine-sulphate solution. This spray solution, not including the cost of preparing it, is almost as expensive as nicotine sulphate solution (1:800 of soap solution). Formula 3A (Table IX), the one recommended against the hop aphid, was found efficient on only the nasturtium aphid, although it was sprayed upon six other species.

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